

FIG. 1a(1)

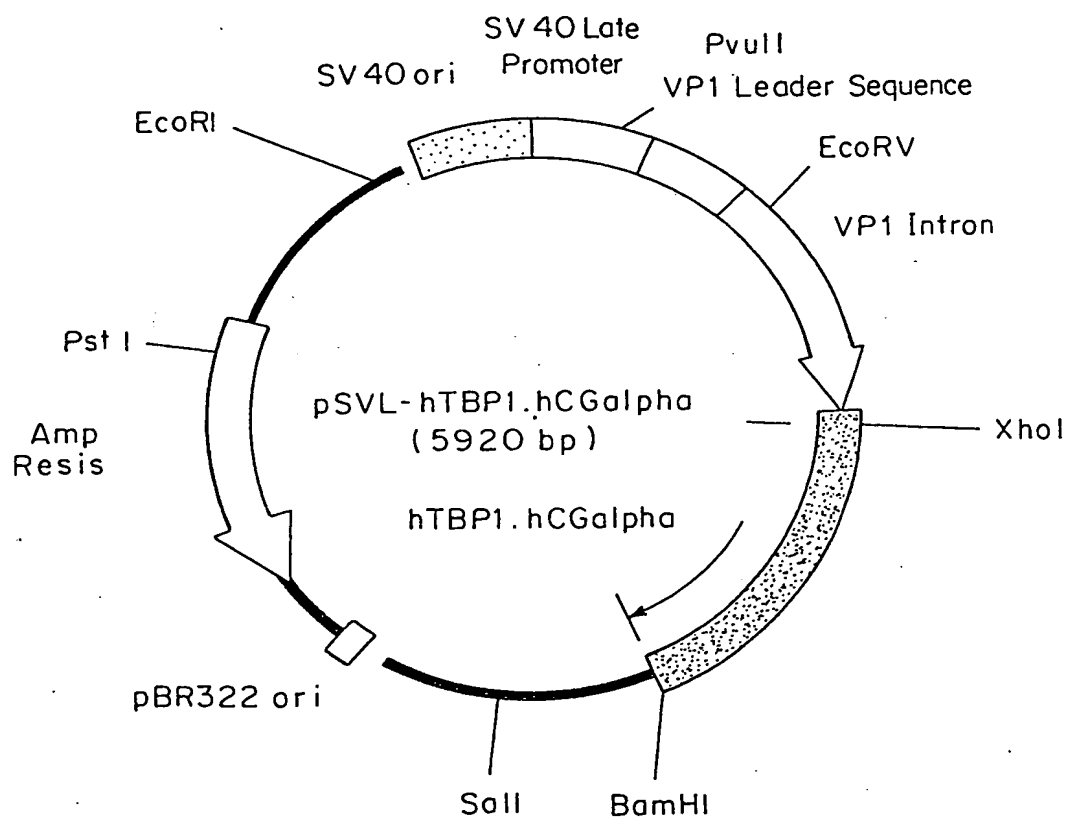


FIG. 1a(1)

FIG. 1a(2)

Xho I hGH Signal Sequence

hGH Intron

TCGAG ATG GCT ACA G GTAAGCGCCCTAAATCCCTTTGGGCACATGTGTCTCTGAGGGGAGAGGAGCGGACCTCTAGATGGACGGGGGCACATAACCTCAGGTTTGGGGTTTCT
 Met Ala Thr

GAATGTAGTATCGCCCATGTAAAGCCAGTATTGGCCCAATCTCAGAAAGCTCTCTGGTCCCTGGAGGGATGGAGAGAGAAAAACAACAGCTCTCTGGACAGGGAGAGTGTCTGTCTTCT

CGGCTCCCTCTGTGGCCCTCTGTGTTCTCCCCAGGC TCC CGG ACG TCC CTG CTC CTG GCT TTT GGC CTG CTG TGC CTG CCC TGG CTT
 Ser Arg Thr Ser Leu Leu Leu Ala Phe Gly Leu Leu Cys Leu Pro Trp Leu

+20 Asp of Processed TBPI

CAA GAG GGC AGT GCC GAT AGT GTG TGT CCC CAA GGA AAA TAT ATC CAC CCT CAA AAT AAT TCG ATT TGC TGT ACC AAG TGC CAC AAA AAA
 Gln Glu Gly Ser Ala Asp Ser Val Cys Pro Gln Gly Lys Tyr Ile His Pro Gln Asn Ser Ile Cys Cys Thr Lys Cys His Lys Gly

ACC TAC TTG TAC AAT GAC TGT CCA GGC CCG GGC GAT ACG GAC TGC AGG GAG TGT GAG AGC GGC TCC TTC ACC GCT TCA GAA AAC CAC CAC CTC
 Thr Tyr Leu Tyr Asn Asp Cys Pro Gly Pro Gly Gln Asp Thr Asp Cys Arg Glu Cys Glu Ser Phe Thr Ala Ser Glu Asn His Leu

AGA CAC TGC CTC AGC TGC TCC AAA TGC CGA AAG GAA ATG GGT CAG GTG GAG ATC TCT TCT TGC ACA GTG GAC CGG GAC ACC GTG TGT GGC TGC
 Arg His Cys Leu Ser Cys Ser Lys Cys Arg Lys Glu Met Gly Gln Val Glu Ile Ser Ser Cys Thr Val Asp Arg Asp Thr Val Cys Gly Cys

AGG AAG AAC CAG TAC CGG CAT TAT TGG AGT GAA AAC CTT TTC CAG TGC TTC AAT TGC AGC CTC TGC CTC AAT GGG ACC GTG CAC CTC TCC TGT
 Arg Lys Asn Gln Tyr Arg His Tyr Trp Ser Glu Asn Leu Phe Gln Cys Phe Asn Cys Ser Leu Cys Leu Asn Gly Thr Val His Leu Ser Cys

Linker

CAG GAG AAA CAG AAC ACC GTG TGC ACC TGC CAT GCA GGT TTC TTT CTA AGA GAA AAC GAG TGT GTC TCC TGT GCC GGT GCT GCC CCA GGT
 Gln Glu Lys Gln Asn Thr Val Cys Thr Cys His Ala Gly Phe Phe Leu Arg Glu Asn Glu Cys Val Ser Cys Ala Gly Ala Ala Pro Gly

+7 Cys of hCG alpha

TGC CCA GAA TGC ACG CTA CAG GAA AAC CCA TTC TCC CAG CCG GGT GCC CCA ATA CTT CAG TGC ATG GGC TGC TGC TCT TCT AGA GCA TAT
 Cys pro Glu Cys Thr Leu Gln Glu Asn pro Phe Ser Gln pro Gly Ala pro Ile Leu Gln Cys Met Gly Cys Cys Phe Ser Arg Ala Tyr

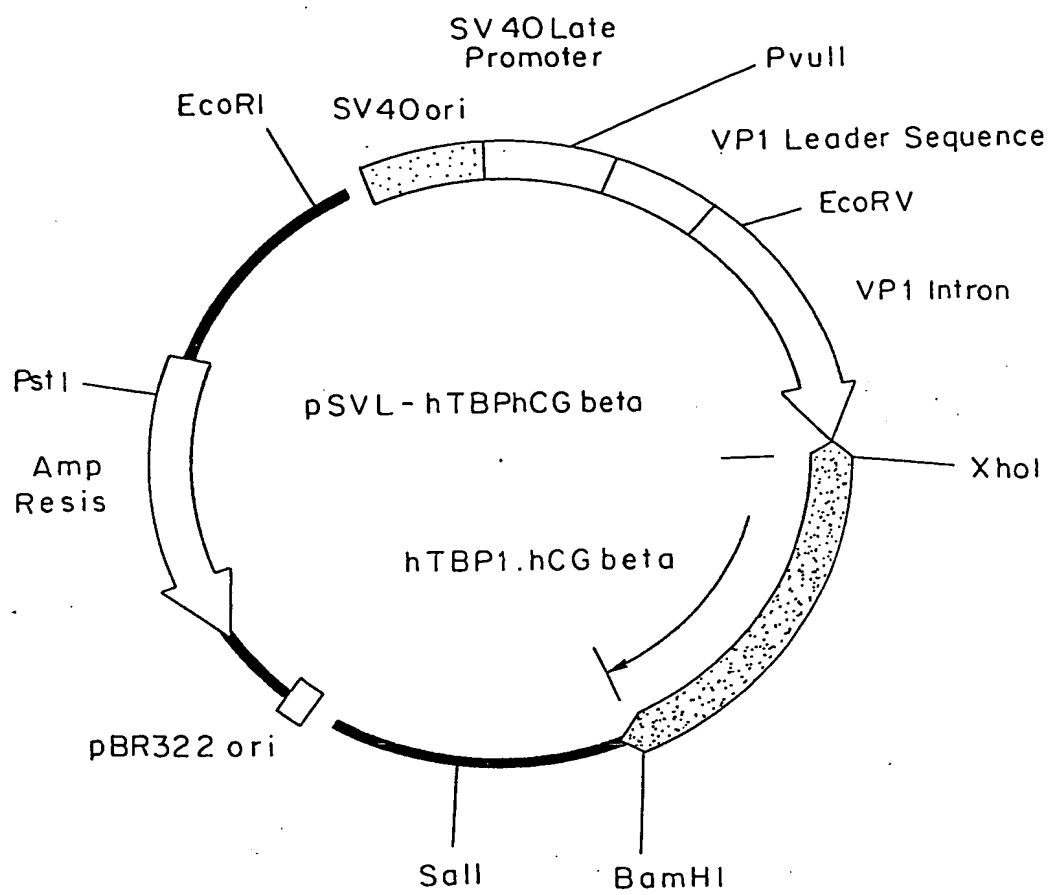
CCC ACT CCA CTA AGG TCC AAG AAG ACG ATG TTG GTC CAA AAG AAC GTC ACT TCA GAG TCC ACT TGC TGT GTA GCT AAA TCA TAT AAC AGG GTC
 Pro Thr pro Leu Arg Ser Lys Lys Thr Met Leu Val Gln Lys Asn Val Thr Ser Glu Ser Thr Cys Cys Val Ala Lys Ser Tyr Asn Arg Val

ACA GTA ATG GGG GGT TTC AAA GTG GAG AAC CAC ACG GCG TGC CAC TGC AGT ACT TGT TAT TAT CAC AAA TCT TAA G

Thr Val Met Gly Gly Phe Lys Val Glu Asn His Thr Ala Cys His Cys Ser Thr Cys Tyr Tyr His Lys Ser ... |

Bam HI

FIG. 1b(1)



00756400 010001

FIG. 1b(2)

hGH Signal Sequence
XhoI
CTCGAG ATG GCT ACA G GTAAGCGCCCTAAATCCCTTTGGGCACAAATGTCCTGAGGGGAGAGTACGACCTGTAGTGGGACGGGGGGCACTAACCCCTCAGGTTTGGG
Met Ala Thr
hGH Intron
GCTTCTGAATGTAGTATCGCCATGTAAGCCCAAGTATTGGCCCAATCTCAGAAAGCTCTGCTGGAGGATGGAGAGAGAAACAAACAGCTCTCTGGAGCAGGAGAGTGCTGAC
CTCTTGTCTCTCGGCTCCCTCTGTGTCCTCTGTTCTGTTCTCCCCAGGC TCC CGG ACG TCC CTG CTG GCT TTT GGC CTG CTC TGC CTG
Ser Arg Thr Ser Leu Leu Ala Phe Gly Leu Leu Cys Leu
+20 Asp of Processed TBPI
CCC TGG CTT CAA GAG GGC AGT GCC GAT AGT GTG TGT CCC CAA GGA AAA TAT ATC CAC CCT CAA AAT AAT TCG ATT TGC TGT ACC
Pro Trp Leu Gln Glu Gly Ser Ala Asp Ser Val Cys Pro Gln Gly Lys Tyr Ile His Pro Gln Asn Ser Ile Cys Cys Thr
AAG TGC CAC AAA GGA ACC TAC TTG TAC AAT GAC TGT CCA GGC CCG GGC CAG GAT ACG GAC TGC AGG GAG TGT GAG AGC GGC TCC TTC ACC
Lys Cys His Lys Gly Thr Tyr Leu Tyr Asn Asp Cys Pro Gly Pro Gly Gln Asp Thr Asp Cys Arg Glu Cys Gly Ser Phe Thr
GCT TCA GAA AAC CAC CAC TGC CTC AGC TGC TCC AAA TGC CGA AAG GAA ATG GGT CAG GTG GAG ATC TCT TCT TGC ACA GTG GAC
Ala Ser Glu Asn His Leu Arg His Cys Leu Ser Cys Ser Lys Cys Arg Lys Glu Met Gly Gln Val Glu Ile Ser Ser Cys Thr Val Asp
CGG GAC ACC GTG TGT GGC TGC AGG AAG AAC CAG TAC CCG CAT TAT TGG AGT GAA AAC CTT TTC CAG TGC TTC AAT TGC AGC CTC TGC CTC
Arg Asp Thr Val Cys Gly Cys Arg Lys Asn Gln Tyr Arg His Tyr Trp Ser Glu Asn Leu Phe Gln Cys Phe Asn Cys Ser Leu Cys Leu
AAT GGG ACC GTG CAC CTC TCC TGC CAG GAG AAA CAG AAC ACC GTG TGC ACC TGC CAT GCA GGT TTC TTT CTA AGA GAA AAT GAG TGT GTC
Asn Gly Thr Val His Leu Ser Cys Gln Glu Lys Gln Asn Thr Val Cys Thr Cys His Ala Gly Phe Phe Leu Arg Glu Asn Glu Cys Val
Linker
+7 Pro of hCG beta
TCC TGT GCT GGT GCT CCA CGG TGC CGC CCC ATC AAT GCC ACC CTG GCT GTG GAG AAG GAG GGC TGC CCC GTG TGC ATC ACC GTC
Ser Cys Ala Gly Ala Gly Pro Arg Cys Arg Pro Ile Asn Ala Thr Leu Ala Val Glu Lys Glu Cys Pro Val Cys Ile Thr Val
AAC ACC AAC ATC TGT GCC GGC TAC TGC CCC ACC ATG ACC CGC GTG CTG CAG GGG GTC CTG CCG GCC CTG CCT CAG GTG GTG TGC AAC TAC
Asn Thr Thr Ile Cys Ala Gly Tyr Cys Pro Thr Met Thr Arg Val Leu Leu Gln Gly Val Leu Pro Ala Leu Pro Gln Val Val Cys Asn Tyr
CGC GAT GTG CGC TTC GAG TCC ATC CGG CTC CCT GGC TGC CCG CGC GTG AAC CCC GTG GTC TCC TAC GCC GTG GCT CTC AGC TGT CAA
Arg Asp Val Arg Phe Glu Ser Ile Arg Leu Pro Gly Cys Pro Arg Gly Val Asn Pro Val Val Ser Tyr Ala Val Ala Leu Ser Cys Gln
TGT GCA CTC TGC CGC CGC ACC ACT GAC TGC GGC GGT CCC AAG GAC CAC CCC TTG ACC TGT GAT GAC CCC CGC TTC CAG GAC TCC TCT
Cys Ala Leu Cys Arg Arg Ser Thr Thr Asp Cys Gly Gly Pro Lys Asp His Pro Leu Thr Cys Asp Pro Arg Phe Gln Asp Ser Ser
TCC TCA AAG GCC CCT CCC AGC CTT CCA AGC CCA TCC CGA CTC CCG GGG CCC TCG GAC ACC CCG ATC CTC CCA CAA TAA
Ser Ser Lys Ala Pro Pro Ser Leu Pro Ser Arg Leu Pro Gly Pro Ser Asp Thr Pro Ile Leu Pro Gln ***

Bam HI

FIG. 2a(1)

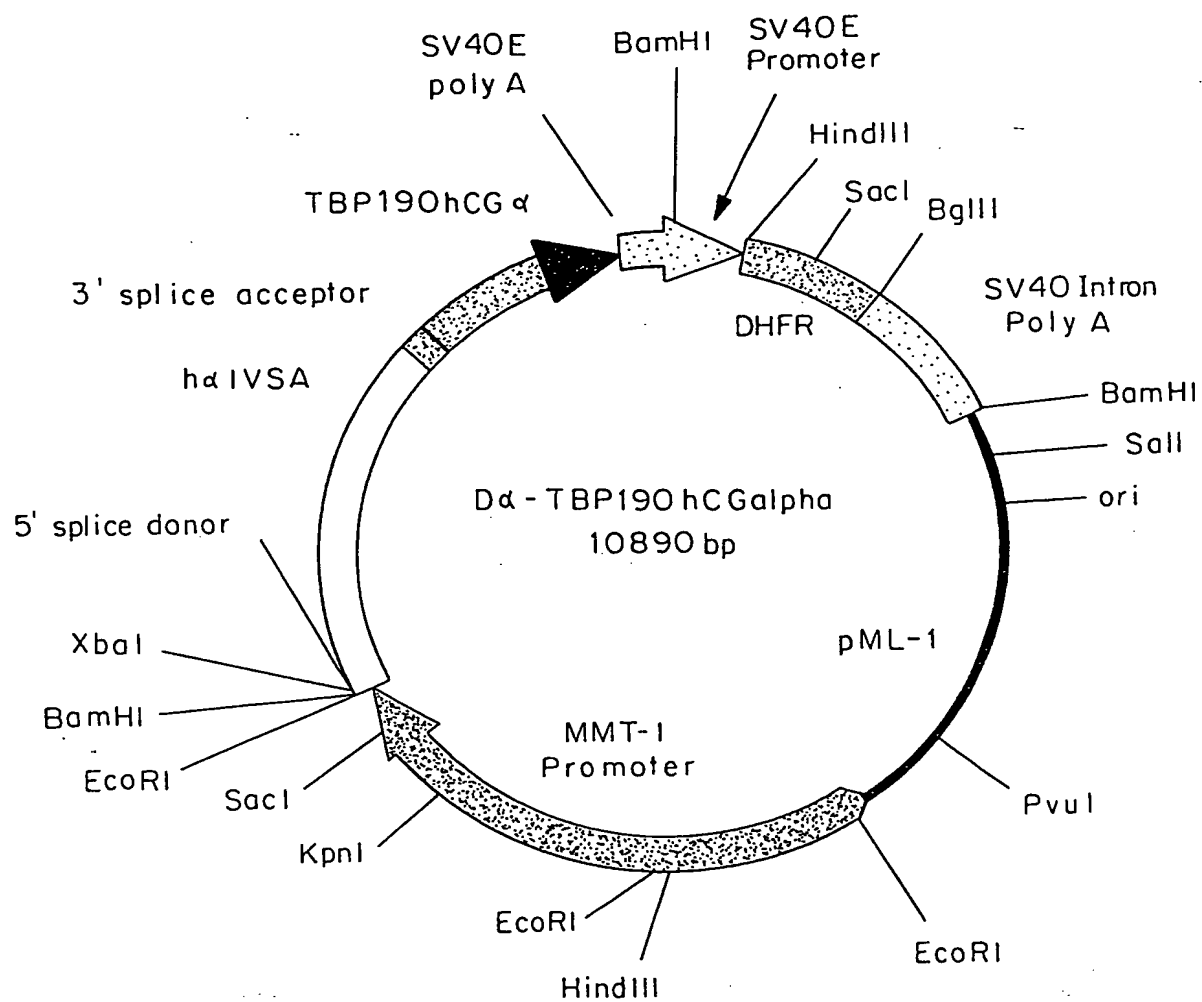


FIG. 2a(1)

TCGAG ATG GCT ACA G
 ▶ Met Ala Thr

GTAAGGCCCTAAATCCCTTTGGGCAACAATGTCTCAGGGGAGAGCGACCTTAGATGGGACGGGGGCACCTAACCTCAGGTTTGGGGTTCT

CGGCTCCCTCTGTGCGCCCTCTGGTTCTCCCCAGGC...TCC CGG ACG TCC CTG CTC CTG GCT TTT GGC CTG CTC TGC CTG CCC TGG CTT

+20 Asp of processed TBPI

CAA CAG GCG AGT GCC GAT AGT GTG TGT CCC CAA GGA AAA TAT ATC CAC CCT CAA AAT AAT TCG ATT TGC TGT ACC AAG TGC CAC AAA GGA

► ACC TAC TTG TAC AAT GAC TGT CCA GGC CCG GGG CAG GAT ACG GAC TGC AGG GAG TGT GAG AGC GGC TCC TTC ACC GCT TCA GAA AAC CAC CTC

AGA CAC TGC CTC AGC TGC TCC AAA TGC CGA AAG GAA ATG GGT CAG GTG ACA GTG GAC CGG GAC ACC GTG TGT GGC TGC
 ▶ Arg His Cys Leu Ser Cys Ser Lys Cys Arg Lys Glu Met Gly Gln Val Glu Ile Ser Ser Cys Thr Val Asp Arg Asp Thr Val Cys Gly Cys

AGG AAG AAC CAG TAC CGG CAT TAT TGG AGT GAA AAC CTT TTC CAG TGC TGC AAT GGC ACC GTG CAC CTC TGC
 ▶ Arg Lys Asn Gln Tyr Arg His Tyr Trp Ser Glu Asn Leu Phe Gln Cys Phe Asn Cys Ser Leu Cys Leu Asn Gly Thr Val His Leu Ser Cys

CAG GAG AAA CAG AAC ACC GTG TGC ACC TGC CAT GCA GGT TTC TTT CTA AGA GAA AAC GAG TGT TCC TGT ACT AAC TGT AAG AAA AGC CTG

► GAG TGC ACG AAG TTG TGC CTA CCC CAG ATT GAG AAT GTT AAG GGC ACT GAG GAC TCA GGC ACC ACA GCC GGT GCT GCT CCA GGT TGC CCA GGT

► GAA TGC ACG CTA CAG GAA AAC CCA TTC TTC CCG GGT GCC CCA ATA CTT. CAG TGC ATG GGC TGC TGC TCT TCT AGA GCA TAT CCC ACT

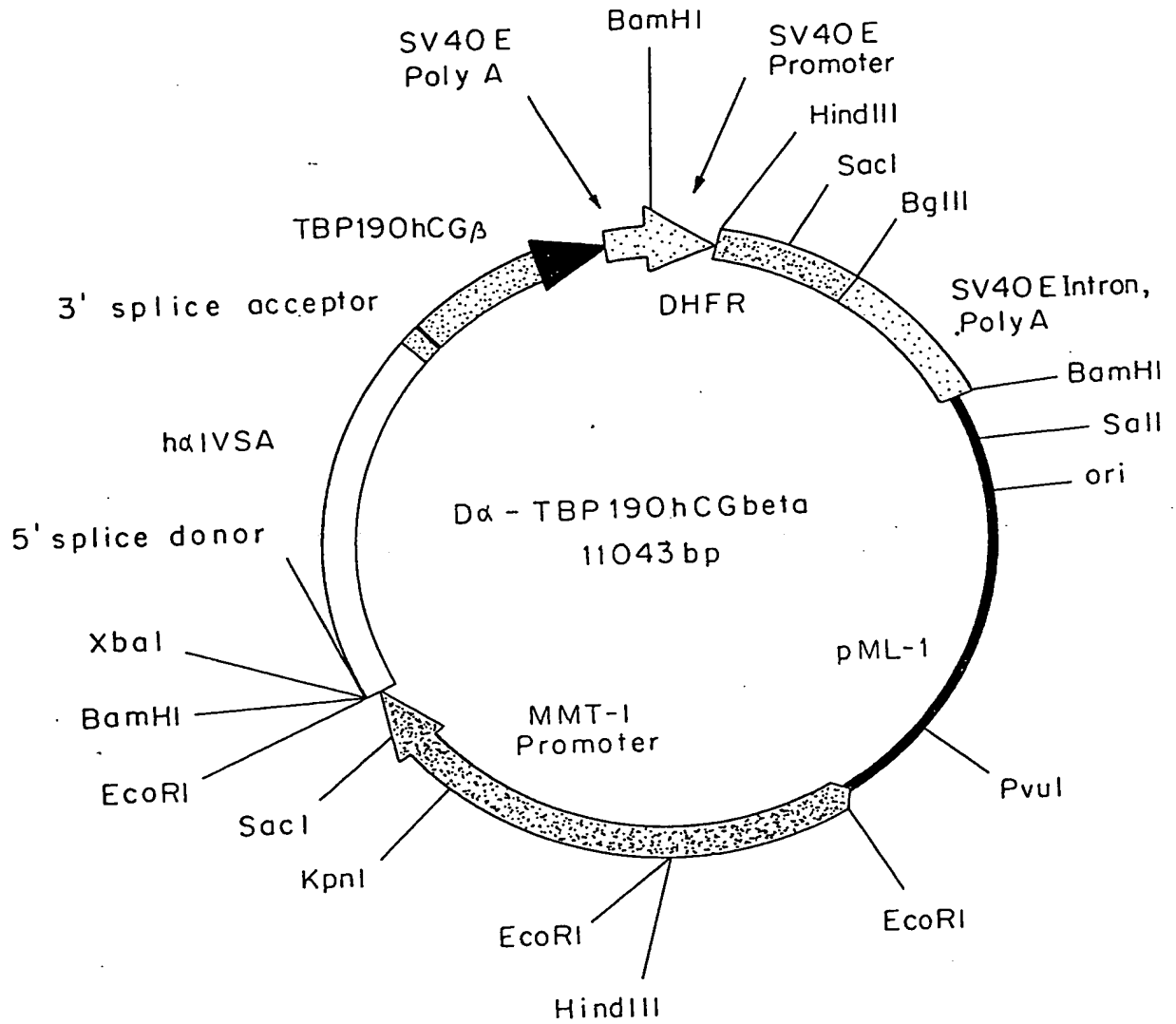
► CCA CTA AGG TCC AAG NAG ACG ATG TTG GTC CAA AAG AAC GTC ACC TCA GAG TCC ACT TGC TGT GTA GCT AAA TCA TAT AAC AGG GTC ACA GTA

ATG GGG GGT TTC AAA GTG GAG AAC CAC ACG GCG TGC CAC TGC AGT ACT TGT TAT TAT CAC AAA TCT TAA GGATCCCTCGAG

Met Gly Gly Phe Lys Val Glu Asn His Thr Ala Cys His Cys Ser Thr Cys Tyr Tyr His Lys Ser ***

Bam HI XhoI

FIG. 2b(1)

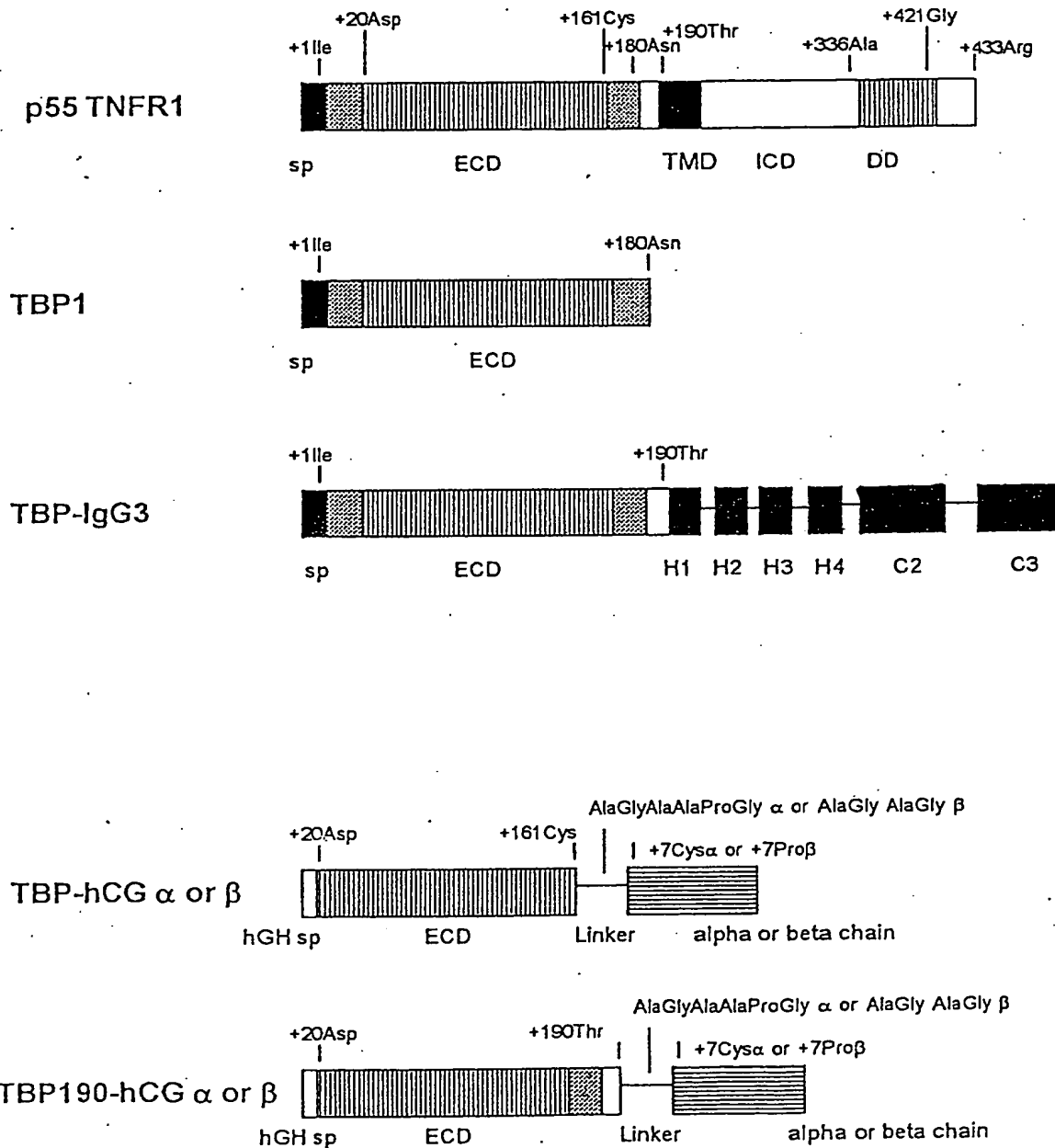


09755185 010901

+7 Pro of beta

FIG. 3

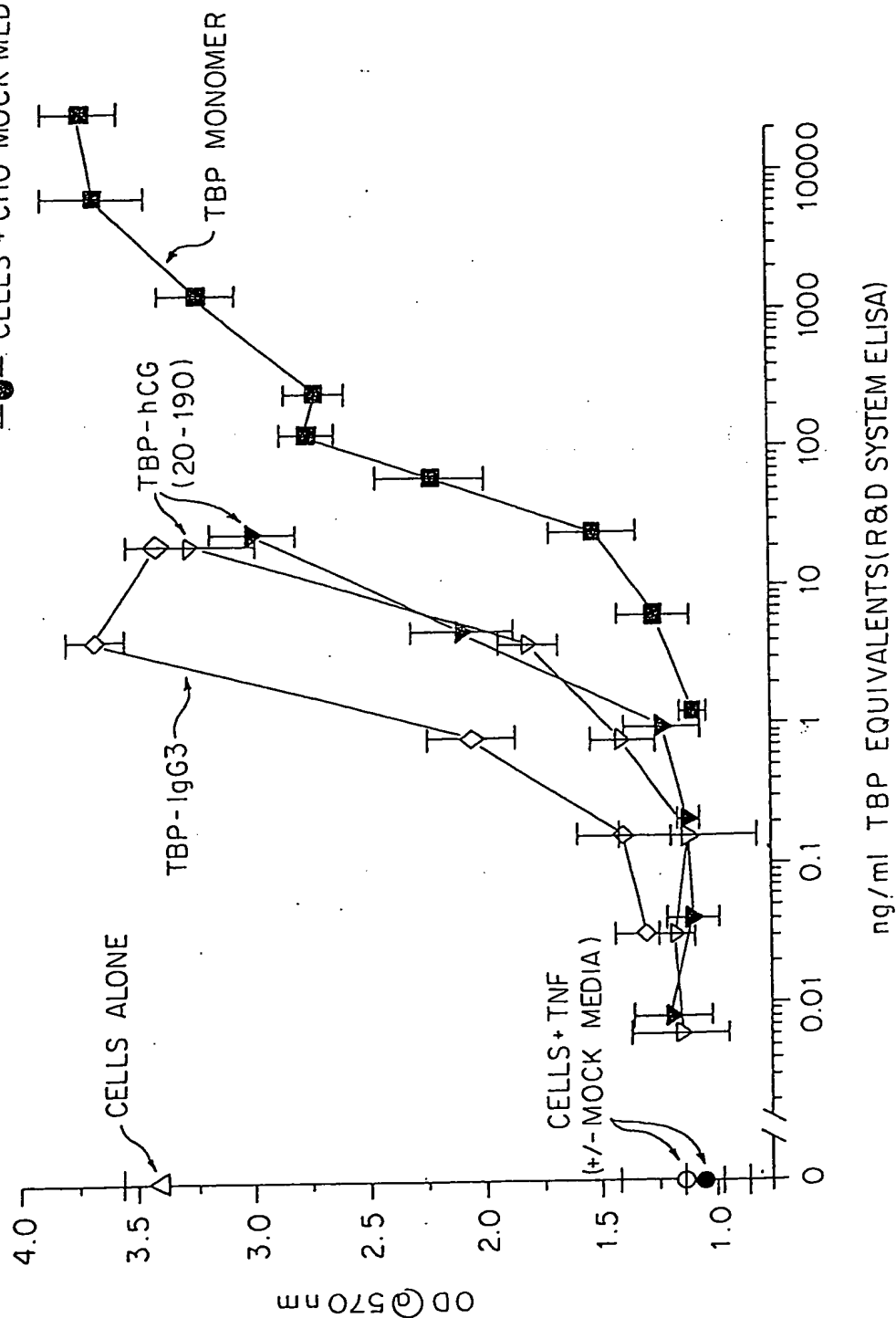
p55 TNFR1, TBP1 and TBP1 FUSION CONSTRUCTS



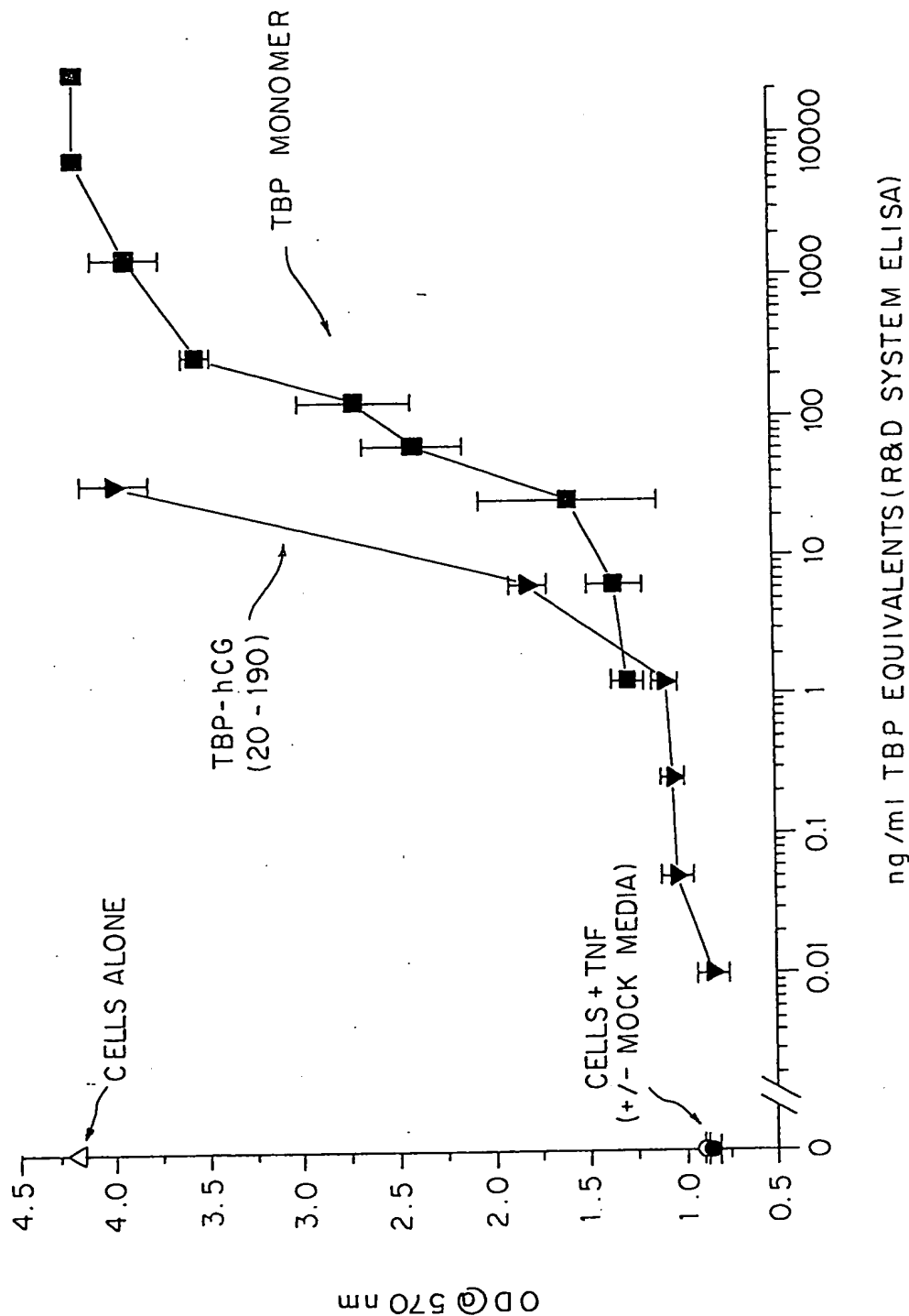
09756486 040904
1000000 9805260

FIG. 4

- 10⁵ CELLS/WELL + 2.5ng/ml TNF α + TBP MONOMER.
- △ CELLS ALONE
- 2.5 ng/ml TNF α (NO TBP)
- ◇— TBP-IgG3 CHO MED + 2.5 ng/ml TNF α
- ▼— TBP-hCG (20-190) CHO MEDIA + 2.5ng/ml TNF α
- ▽— TBP-hCG (20-190) CHO MEDIA + 2.5 ng/ml TNF α
- CELLS + CHO MOCK MEDIA + 2.5 ng/ml TNF α



- 10⁵ CELLS / WELL + 2.5 ng/ml TNF α + TBP MONOMER
- △ CELLS ALONE
- CELLS + 2.5 ng/ml TNF α (NO TBP)
- ▼— CELLS + TBP-hCG (20-190) COS7 MED + 2.5 ng/ml TNF α
- CELLS + COS7 MOCK TRANSFECTANT MEDIA + 2.5 ng/ml TNF α



—■— 10⁵ CELLS / WELL + 2.5 ng/ml TNFα + TBP MONOMER

△ CELLS ALONE

○ CELLS + 2.5 ng/ml TNFα (NO TBP)

—▽— CELLS + PURIFIED TBP-hCG (20-161)

FIG. 6

